Proposed Title: Drinking Water Contamination in NC: Water Use, Human Health and Going Beyond GenX

BACKGROUND AND MOTIVATION. Safe and clean drinking water in the United States is no longer a guarantee. The complex nature of water acquisition and governance in the US creates substantial challenges for water protection, and communities, states, and consumers bear the social and economic burden when drinking water sources are contaminated. Preventing source water contamination is less costly than remedying its effects; however, the majority of water contamination events are only brought to light after health concerns are suspected and remediation is required. The most relevant threat to public drinking water in the United States today is a class of 3,000+ chemicals called poly and perfluoroalkyl substances (PFASs) that have been released into the environment since the 1950s[1]. Referred to as "Forever Chemicals" due to their long chains of carbon bonded to multiple fluorine atoms, PFASs are durable and long-lived chemicals that are resistant to degradation.

PFAS chemicals are widely used in industrial processes and consumer products globally. Because they are able to repel both moisture and oil they are commonly used in consumer products as stain, water and grease repellents in carpets, upholstery, clothing and even nonstick coatings. As such, PFASs are pervasive in our homes, schools and work environments. These compounds are also heavily used in fire-fighting foams, and as a result, are often detected at high levels in surface water and drinking water near military bases, and airports that use these foams for fighting fires and often for training purposes. Due to their ubiquitous use in products, PFAS exposure is widespread, and more than 95% of the US population has PFAS chemicals in their bodies[2]. Drinking water is considered a primary source of exposure, and research is now uncovering PFASs in water utilities all over the country, sparking concerns about health impacts in affected communities **(Fig. 1)**. A recent study in West Virginia concluded that exposure to PFOA, one type of PFAS, was associated with several adverse health outcomes, including thyroid disease, testicular cancer, kidney cancer, ulcerative colitis, and pregnancy-induced hypertension[3]. Additional research on PFAS suggests that exposures in pregnant women are associated with low birthweight and preterm birth[4,5]. Preterm birth accounts for 34% of all infant deaths in the US and costs the US healthcare system $26.2 billion dollar annually. While most babies survive, those born too early are at increased risk of chronic health conditions throughout their lifetimes, including respiratory and metabolic disorders.

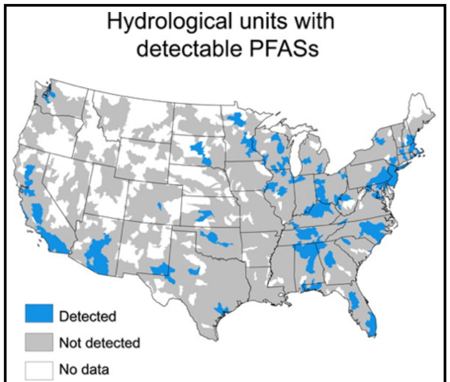

Figure 1. PFAS are detected in water samples from across the U.S. (from Hu et al. 2016.)

PFAS chemicals are now a North Carolina (NC) problem. These chemicals first made local headlines in June 2017 when a North Carolina news stations reported contamination of drinking water wells with "**GenX**" in New Hanover and Brunswick counties[6]. GenX is the nickname given to a new type of PFAS chemicals that is manufactured at a local chemical company (Chemours) in Bladen county. It has been dumped into the Cape Fear River Basin for decades, contaminating the drinking water source for hundreds of thousands of NC residents. By 2018 the NC legislature appropriated several million dollars to support academic researchers at UNC Chapel Hill, NC State, ECU and Duke University to test all surface waters across the state for PFASs and investigate remediation strategies. Preliminary data from surface water collections demonstrated that PFASs are present in the tap water of residents from Chapel Hill, Durham, Pittsboro, Cary and Raleigh, and notably, **PFAS levels in Pittsboro are similar to the GenX levels measured along the Cape Fear River basin**.

We hypothesize that there are several point sources of PFAS contamination to local drinking water sources in the NC Triangle region, and furthermore, we hypothesize that these exposures may be related to poorer birth outcomes (e.g. low birthweight) in resident populations. Despite the aforementioned broad documentation of PFAS contamination in NC surface water, **no funding was included by the NC legislature to evaluate the impact of these chemical exposures on health outcomes of the NC population nor hydrodynamic modeling to evaluate sources and water use policies in the Research Triangle**. As such, we propose a study that will *extend the scope of projects funded by the NC legislature* to Heather Stapleton (NSOE) and P. Lee Ferguson (Pratt) to further investigate PFAS issues in Duke's backyard. Our "Beyond GenX" Collaboratory will 1) construct a hydrodynamic model to fully characterize the point source(s) of PFAS contamination in the NC piedmont region and identify solutions to reduce exposure of residents to contaminated water, 2) define the

relationship between environmental and biological (e.g. human serum) PFAS levels, 3) establish the association between PFAS levels and preterm birthrates, and 4) integrate environmental and human health knowledge into actionable management and policy outcomes that preserve water quality. To meet these goals we will leverage Duke expertise in environmental chemistry, hydrology, water policy, exposure science, toxicology and epidemiology, and engage a local community organization, the Haw Riverkeeper, to address community concerns. This proposed study targets two of the three strategic priorities of the Together Duke initiative: water resources and population health, and would engage a group of master's and doctoral students in a current problem located close to our University.

Preliminary Data. In Sept. 2018, Drs. Stapleton and Ferguson received funding from the NC legislature to investigate PFAS levels in NC surface waters (Ferguson) and in drinking water (Stapleton). Dr. Stapleton is also investigating the efficacy of in-home filters in removing the PFASs from tap water. Preliminary data indicate PFASs are detectable in all drinking water sources in the Triangle (**Fig. 2**), and levels in Pittsboro are particularly elevated compared to other cities (i.e. Pittsboro PFAS levels are 30 times greater than those in Durham). Preliminary data demonstrate that PFAS chemicals are *not removed* by standard water filters (e.g. pitcher, refrigerator filters, etc.) but that reverse osmosis (RO) filters are capable of effectively removing all PFASs from water. However, the high cost of installing a home RO system is likely a major obstacle for a majority of families.

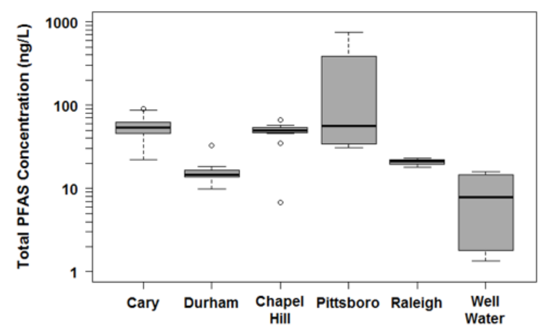

Figure 2. PFAS levels measured in tap water from different water utilities in the Triangle vary considerably.

As part of an NIH funded study, Dr. Stapleton's research group also investigated the associations between maternal PFAS exposure during pregnancy, and infant's weight and gestational age at birth among women living in Durham, NC. Women with the highest levels of PFAS exposure gave birth to babies an average of 15 days earlier than women with the lowest levels of exposure ($p<0.01$). This is particularly concerning because babies born early and with low birth weight are at higher risk of for chronic diseases later in life (e.g. heart disease, diabetes). Given that levels of PFAS exposure as substantially higher in other Triangle drinking water systems, it is critical to evaluate birth outcomes in additional communities.

Research & Project Goals. Preliminary data highlight an urgent need to leverage the NC legislative fund supporting PFAS monitoring across the state of NC and determine if communities with higher PFAS levels in drinking water (like Pittsboro) are grappling with higher probabilities of adverse health outcomes. Furthermore, it's important that we understand the sources of PFASs to these water sources, understand the factors influencing their fate and transport, and work with impacted communities and water utilities to identify optimum solutions to reduce exposure. This interdisciplinary study will accomplish these goals (**Fig. 3**). This project will include a team of faculty and researchers from the Nicholas School of the Environment, the Pratt School of Engineering, the Nicholas Institute for Policy Solutions, and the Department of Obstetrics & Gynecology. It will further provide critical hands-on training to a group of master's students in the Nicholas Master's of Environmental Management program and will engage students in a current and local problem related to water resources and health. Below we outline five different research aims supported by this project and describe their integration.

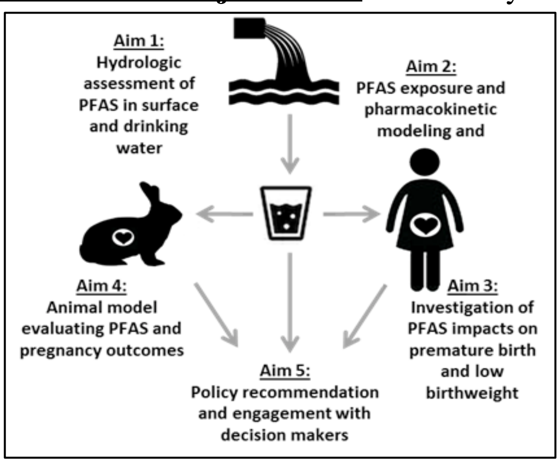

Figure 3. Overview schematic of proposed study.

Specific Aim 1: *Construct a hydrodynamic model of the Haw River/Jordan Lake area to predict PFAS drinking water levels in Pittsboro and Cary/Apex* (Lead Investigator: Dr. Kateri Salk; Co-investigator: P. Lee Ferguson). Preliminary data generated by Dr. Stapleton's research group demonstrate that PFAS concentrations in the Haw

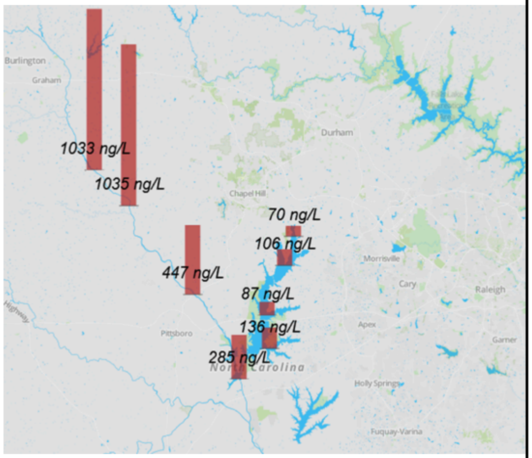

Figure 4. Total PFAS concentrations measured along the Haw River and Jordan Lake in July 2018.

River (which provides drinking water in Pittsboro) are negatively correlated with flow rates, and PFAS concentrations decrease moving downstream from the city of Burlington (**Fig. 4**). The patterns of PFAS concentrations over time and space indicate point source discharges from wastewater treatment plants (WWTPs) followed by a dilution moving downstream and under high flow. The objectives for this aim are to (1) determine the magnitude of PFAS discharges from WWTPs in Burlington, (2) define empirical relationships between Haw River flow rates and PFAS concentrations, and (3) model the hydrodynamics and contaminant transport of Jordan Lake and its tributaries. Data will be gathered from the city of Burlington WWTPs, pollutant discharge permits, and streamflow monitoring sites to determine the proportion of flow in the Haw River attributed to point source discharges. Water samples for PFAS analysis will be collected in several locations in the Haw River and Jordan Lake, moving downstream from Burlington WWTPs. To better define the empirical relationship of flow rates and PFAS concentrations, samples will be collected monthly to capture baseflow conditions (year 1) and intensively during storm events (years 1-2). Additional well-studied contaminants associated with WWTPs (e.g., artificial sweeteners) will be investigated as potential proxies to predict PFAS concentrations. We will apply a hydrodynamic model for Jordan Lake and its tributaries, which was previously validated for nutrients but will be newly applied to PFAS chemicals. The complex hydrology of this system will be characterized to predict the fate of PFAS chemicals as they are transported through rivers and into reservoirs where drinking water intakes are located. Completion of this aim will allow us to generate predictions that will allow municipalities to anticipate periods of high PFAS concentrations and to alter their water sources and/or recommendations to citizens accordingly.

Specific Aim 2: *Examine the relationship between drinking water levels of PFAS and serum levels of PFAS in residents consuming water with elevated PFAS levels* (Lead Investigator: Dr. Heather M. Stapleton; Co-Investigator: P. Lee Ferguson). Drinking water is believed to be the primary source of exposure to PFAS chemicals in the general population. Numerous studies have reported on PFASs in drinking water nationwide, and studies suggest that levels are higher in drinking water with proximity to airports, military bases and waste-water treatment plants. Due to health concerns regarding one particular PFAS (PFOA), researchers constructed a one-compartment pharmacokinetic model to predict serum levels based on concentrations in drinking water, average water consumption rates, partitioning to the blood stream, and persistence in the human body (half-life~ 2.3 years). However, this model has only been used to predict human serum levels of PFOA, and little research has been conducted to determine if this model can be applied to other PFAS chemicals. Given that the PFASs present in the Haw River are not dominated by PFOA, it is important that we investigate the relationship between drinking water levels and human serum levels for other types of PFAS chemicals. Here, we propose to recruit a cohort of approximately 30 participants residing in Pittsboro, NC to assist with this study. Participants will be asked to provide two drinking water samples and two serum samples, two months apart, during which they will keep a diary of their drinking water habits. Water and serum will be analyzed for PFASs and used in the model validation and optimization for PFAS chemicals. These measurements will allow us to model the body burdens of PFAS in residents receiving Pittsboro drinking water.

Specific Aim 3: *Determine if higher levels of PFASs in drinking water are associated with an increased risk of premature birth or low birthweight*. (Lead Investigator, Dr. Kate Hoffman; Co-Investigator: Liping Feng). Exposure to PFASs has been associated with decreased fetal growth and an increased risk of premature birth; however, results of human studies are inconsistent. Prior studies have largely focused on perfluorooctane sulfonate (PFOS) and perfluorooctanoate (PFOA), potentially missing important associations with other PFAS and failing to account for the impacts of exposure to multiple PFAS simultaneously. Disparities in exposure across central North Carolina, as a result of differing levels of PFAS in municipal drinking water systems, provide a unique opportunity to evaluate exposure to PFAS mixtures in relation to birth outcomes in a large

population. The objectives of this aim are to (1) couple measurements of PFAS in municipal drinking water systems (Aim 1) and pharmacokinetic models (Aim 2) with residential history information to estimate gestational exposure to PFAS and (2) estimate associations between gestational exposure to PFAS and birth outcomes (i.e., birthweight and gestational age). In order to meet these objectives, birth certificate data for all children born in NC will be obtained from the NC Department of Health and Human Services (official IRB and DHHS approval for this work have been granted). This aim is expected to contribute an assessment of the potential impacts of a broad range of PFASs on birth outcomes and will provide information on potential health impacts in communities with contaminated drinking water. Using advanced statistical methods we will identify the PFASs having the largest impact on children's growth and development which may provide opportunities for target interventions. In addition, these data could have a substantial public health impact given that even small changes in birth weight and gestational timing have been associated with long-term adverse health risks.

<u>Specific Aim 4:</u> *Determine whether maternal exposure to PFASs impacts placental function and pregnancy outcomes in a rabbit model* (Lead Investigator, Dr. Liping Feng; Co-Investigator Heather Stapleton). Throughout pregnancy, the developing fetus is entirely dependent on the placenta to deliver nutrients, remove wastes and other avenues of support. A growing body of science now suggests that health outcomes in adulthood are linked to stressors experienced in the womb. The rapidly developing placenta and fetus are vulnerable and adversely affected by environmental toxicants. We have shown that one of the PFAS compounds influences placental function in a rabbit model, which may contribute to low birth weight and early birth. In this research aim, we propose to further example the cause-and-effect relationship between PFAS exposure and low birthweight and early birth, using exposures that are similar to exposures experienced in Pittsboro, NC. Rabbits are chosen for these experiments because their placental structure and functions are the closest to humans' outside of primates. Rabbits will be randomly divided into 5 groups (8 rabbits/group) and exposed to PFASs via their drinking water, similar to how people are exposed. The treatment groups are: (1) Control (i.e. no PFAS exposure), (2) solvent control, (3) low dose PFAS mixture, (4) high dose PFAS mixture, and (5) drinking water from Pittsboro. Experimental protocols are designed to start PFASs exposure 100 days (mimicking chronic exposure) before mating until pregnancy day 26, immediately before delivery (gestation length is ~31 days). Rabbits have an average of 8-10 kits per litter, thus we expect that at least 320 placentas and kits will be monitored. Body weights will be recorded weekly. Sample collections and procedures will be performed on the schedule shown in **Table 1**. Outcomes to be examined are also summarized in **Table 1.**

Table 1. Sample collection and outcomes examination

Day	Sample collection and procedures	outcomes
-100	Maternal blood samples	Measurement of PFASs
0	Maternal blood samples and Mating	Measurement of PFASs, cytokines, hormones
16	Doppler and maternal blood samples	Fetal body and head parameter; placental biometry; blood pressure; Measurement of PFASs, cytokines, hormones Observation of gestational length (GA)
26	Doppler, maternal and fetal blood samples, placental and fetal tissues	Fetal body and head parameter; placental biometry; blood pressure Measurement of PFASs, cytokines, hormones, GA Placenta pathology and quantification of PFASs; Cytokines and hormone measurements; Epigenetic assays

<u>Specific Aim 5:</u> *Identify best practices for local government responses to PFASs in water supplies* (Lead Investigator, Kay Jowers; Co-Investigator Kateri Salk). Cary, Apex, and Pittsboro all withdraw water from within the Jordan Lake system. In order to provide these communities with policy recommendations that incorporate the PFAS science, we aim to *assess the existing legal and policy mechanisms to understand when municipal water utilities should or could withdraw water from different sources and when to communicate with citizens regarding unregulated contaminants in water supplies*. We will engage with law students to review *the current legal and policy landscape that governs disclosure of emerging contaminants*, including disclosure requirements for the municipal water utility to disclose to federal or state agencies as well as for disclosure to the using and consuming public. Using interviews with NC communities who have been dealing with GenX in their water supplies, we will be able to understand the mistakes and successes associated information and water dissemination, which may provide recommendations for local government policy approaches to emerging contaminants. These best practices will then be used to aid in the dissemination of the results from Aims 1-4.

By achieving this aim, we will generate policy recommendations for local governments drawing water from the Haw River and Jordan Lake for their drinking water sources. These recommendations will allow municipalities including Pittsboro and Cary/Apex to decide when to withdraw water from other sources and how best to communicate with citizens regarding the risks of exposure to PFASs in water supplies.

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